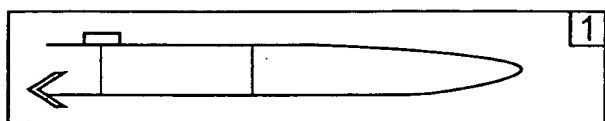PUTATIVE C-PEPTIDE CONTAINING IMPURITIES. ALL OF THE EXAMPLES SHOW
"INSULIN C-PEPTIDE LIKE IMMUNOREACTIVITY"

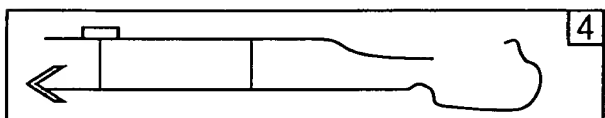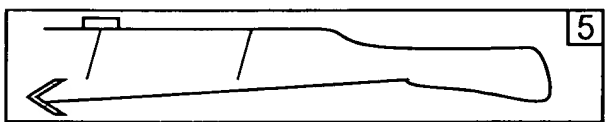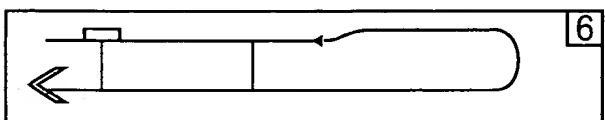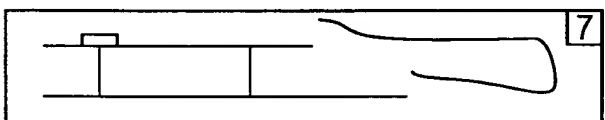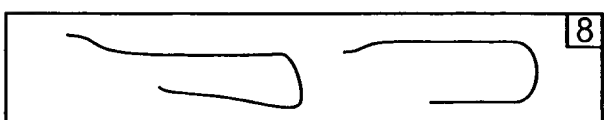|	
[1]	PREPROINSULIN WITH OR WITHOUT PRE-SEQUENCE. MODEL TEST COMPOUND: PURIFIED PPI
[2]	PREPROINSULIN WITH OR WITHOUT PRE-SEQUENCE, CLEAVED AT THE ACID LABILE DP SITE. MODEL TEST COMPOUND: PURIFIED PPI CLEAVED WITH ENDO ASP-N AT THE EDP SITE.
[3]	PREPROINSULIN WITH OR WITHOUT PRE-SEQUENCE, UNPROCESSED AT THE N-TERMINAL BORDER OF A-CHAIN.
[4]	PREPROINSULIN WITH OR WITHOUT PRE-SEQUENCE, UNPROCESSED AT THE C-TERMINAL BORDER OF B-CHAIN. MODEL TEST COMPOUND: HIA2 PPI
[5]	INCORRECTLY FOLDED OR UNFOLDED PREPROINSULIN WITH OR WITHOUT PRE-SEQUENCE. MODEL COMPOUND: PURIFIED PPI WITH REDUCED S-S BONDS AND ALKYLATED CYSTEINES.
[6]	PREPROINSULIN OF HIA2 IT CAN BE USED AS A MODEL TEST COMPOUND FOR 4.
[7]	ISOLATED MONKEY C-PEPTIDE FROM HI OR MUTATED C-PEPTIDE FROM HIA2 IN THE PRESENCE OF CORRECTLY PROCESSED INSULIN.
[8]	ISOLATED C-PEPTIDES FROM HUMAN INSULIN. MODEL TEST COMPOUNDS TO CHECK INFLUENCE FROM DEVIATIONS IN SEQUENCE OR AMINO ACID COMPOSITION.
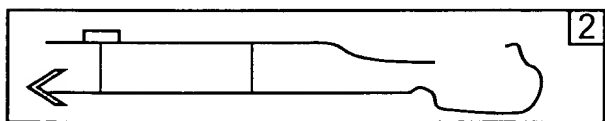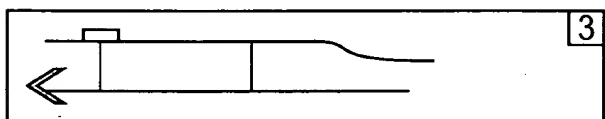
FIG. 1A

EXPLANATIONS

_____ ▢ _____   = A-CHAIN OF INSULIN

_____   = B-CHAIN OF INSULIN
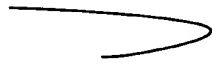
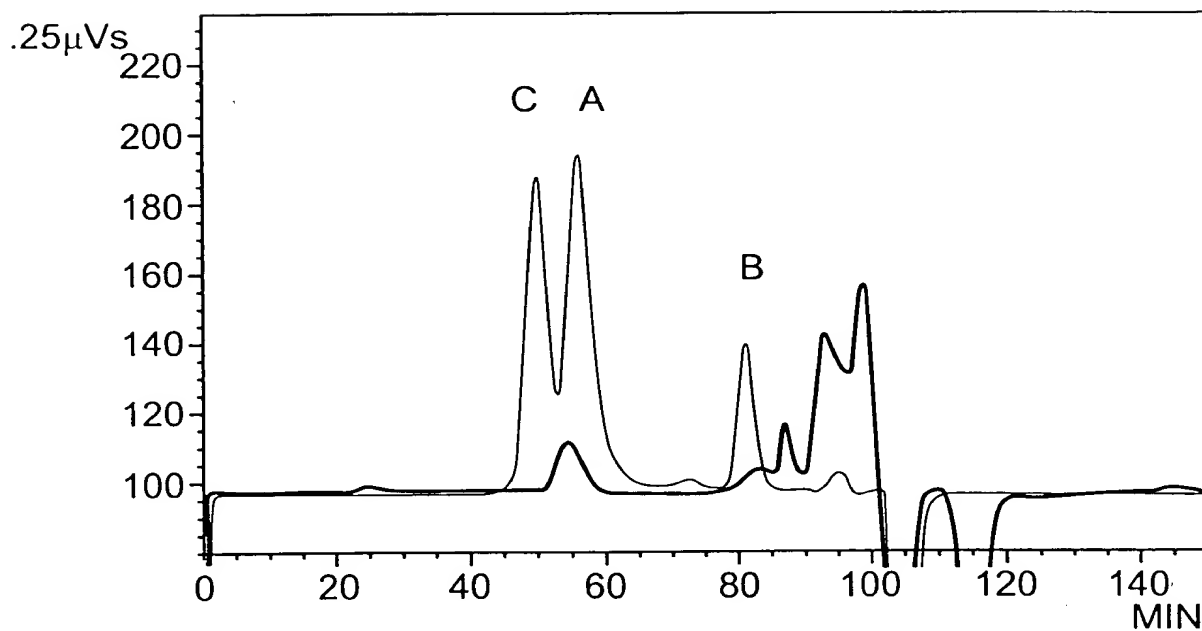⟍_____⟍   = C-PEPTIDE

≪   = PRESEQUENCE OF RECOMBINANT
        INSULINS

| = COVALENT BOND BETWEEN "SH"
       OF CYSTEINES

FIG. 1B

.25µVs

ADC1 A, SIGNAL FROM PC LOOP (5\SDPE_008.D)

ADC1 A, SIGNAL FROM PC LOOP (5\SDPE_031.D)

FIG. 2

ILLUSTRATION OF 6 DIFFERENT INSULIN C-PEPTIDE
STANDARD CURVES AS OBTAINED IN THE BEAD ASSAY
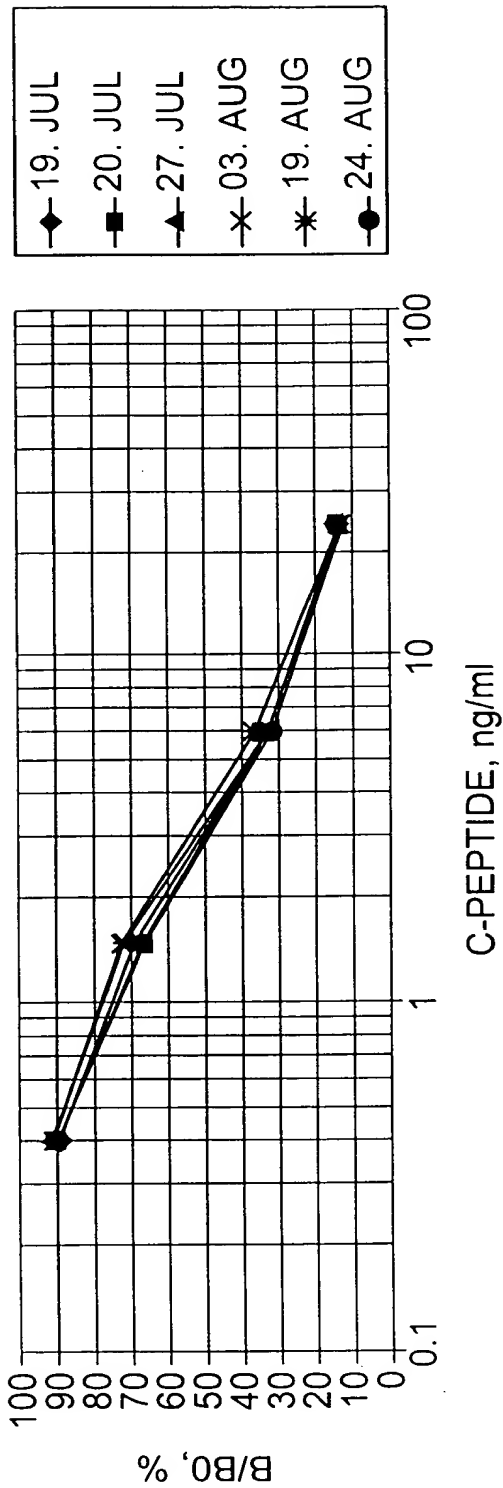
19. JUL
20. JUL
27. JUL
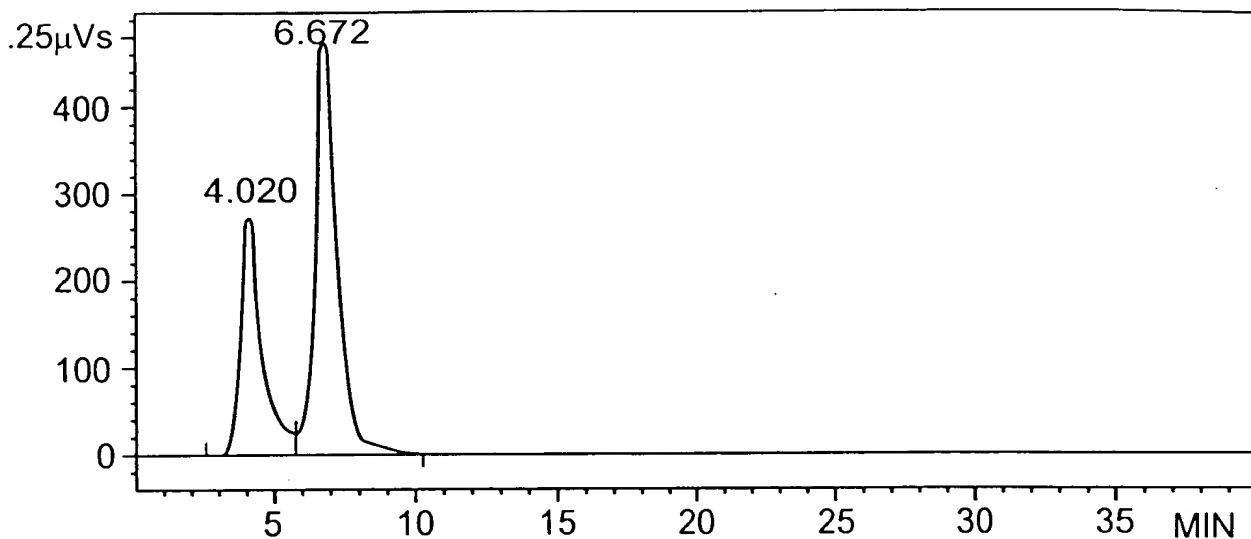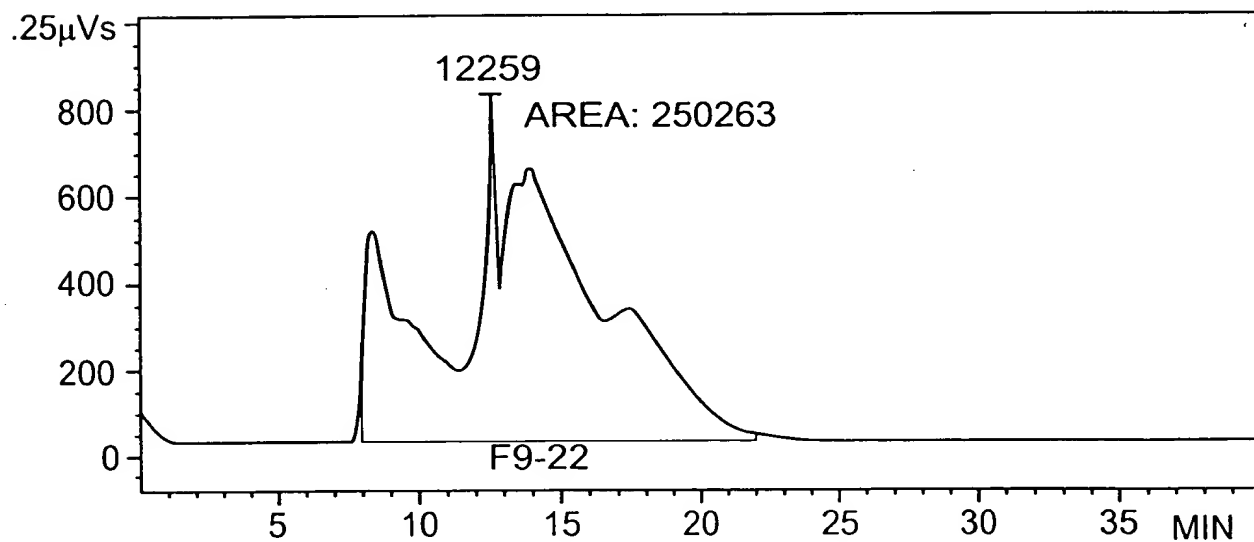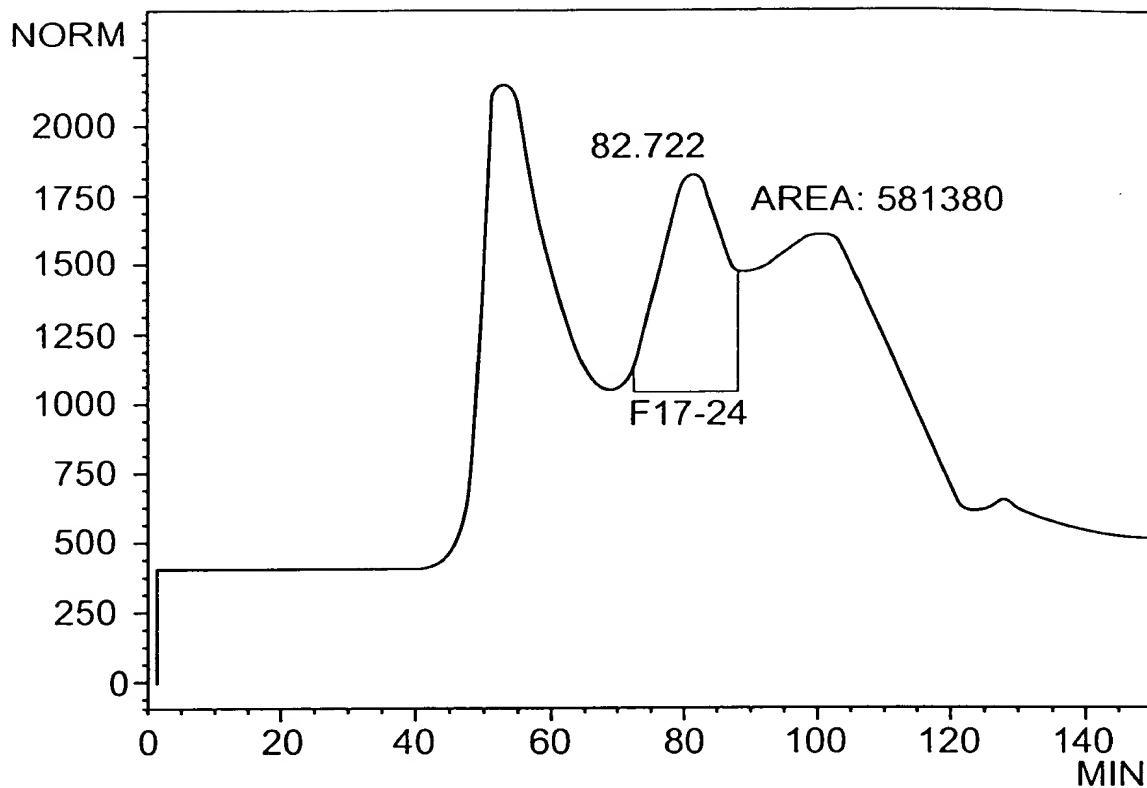03. AUG
19. AUG
24. AUG

C-PEPTIDE, ng/ml

B/B0, %

FIG. 3

ADC1 A, SIGNAL FROM PC LOOP (4\AZL_109.D)

FIG. 4A
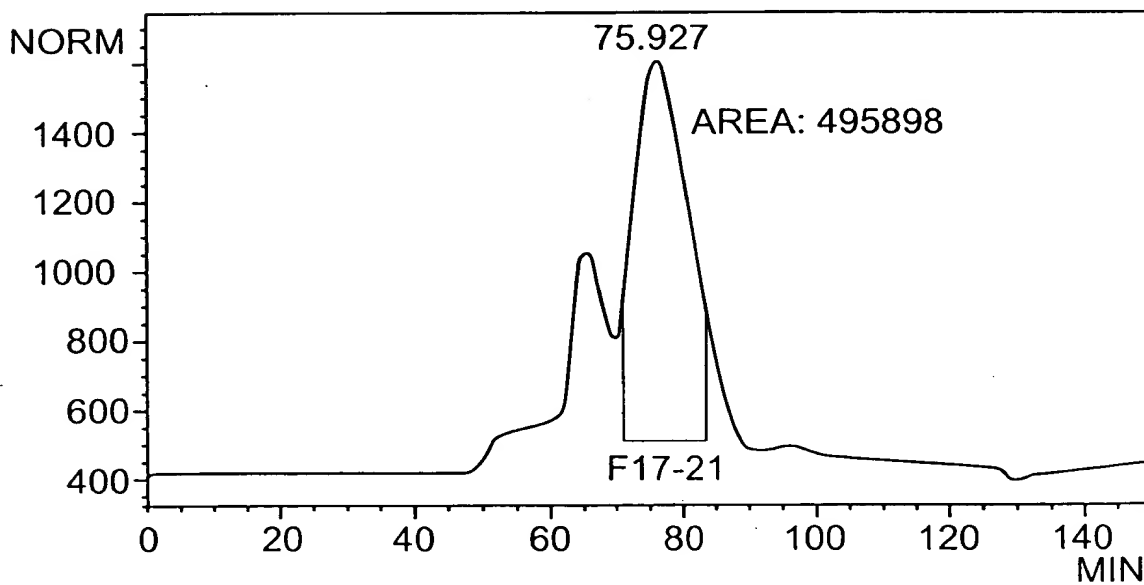

ADC1 A, SIGNAL FROM PC LOOP (4\AZL_108.D)
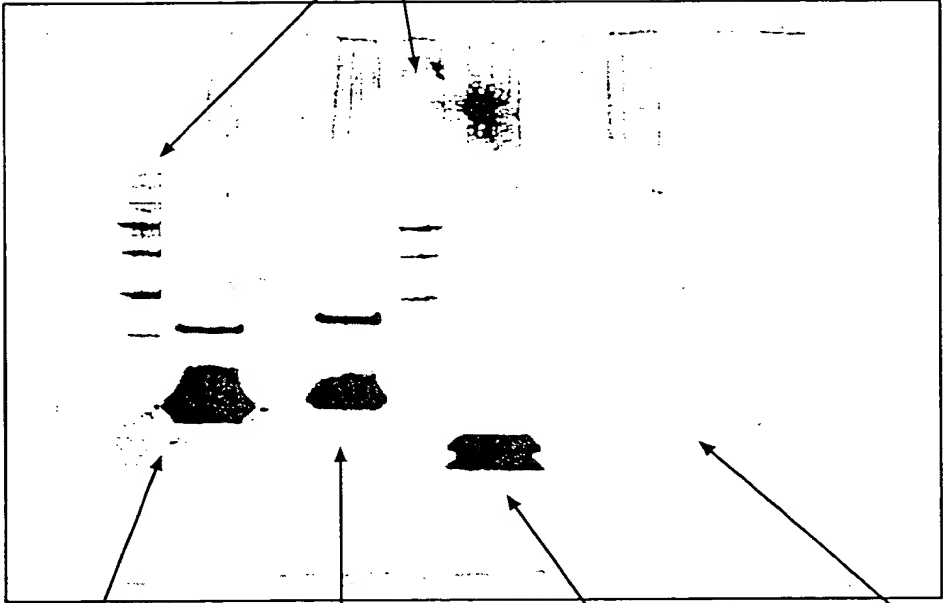
FIG. 4B

ADC1 A, SIGNAL FROM PC LOOP (6\SD2_652.D)

FIG. 5

ADC1 A, SIGNAL FROM PC LOOP (6\SD2_656.D)

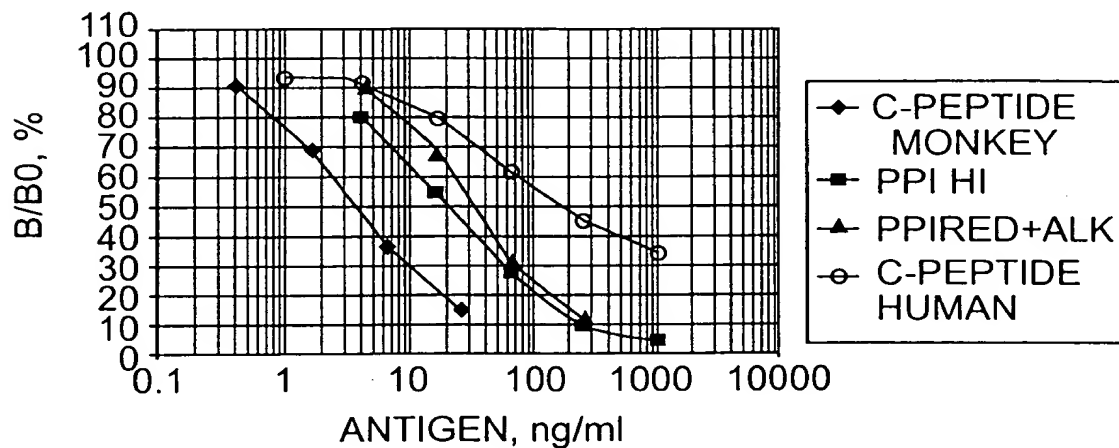*FIG. 6*

MARKER LINES

HI PPI   HI PPI   HI PPI   HI PPI
                REDUCED /   DIGESTED   REDUCED/
                ALKYLATED   WITH ENDO   ALKYLATED
                                 ASP-N   DIGESTED
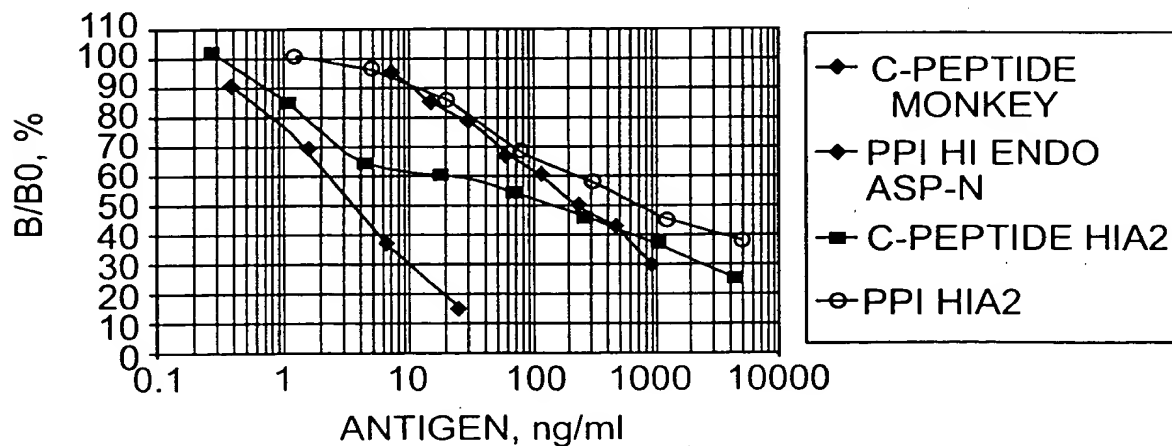   WITH ENDO
                                                 ASP-N

FIG. 7

ANALYSIS OF DIFFERENT CONTROL ANTIGENS USING
THE COATED BEAD CHEMILUMINESCENCE ASSAY

- ◆ C-PEPTIDE MONKEY
- ■ PPI HI
- ▲ PPIRED+ALK
- ○ C-PEPTIDE HUMAN

FIG. 8

ANALYSIS OF DIFFERENT CONTROL ANTIGENS USING
THE COATED BEAD CHEMILUMINESCENCE ASSAY

- ◆ C-PEPTIDE MONKEY
- ◆ PPI HI ENDO ASP-N
- ■ C-PEPTIDE HIA2
- ○ PPI HIA2

FIG. 9